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δ18O analysis of individual carbohydrates - a new method for GC-pyrolysis-IRMS

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Measuring the oxygen isotopic composition (δ18O) of various plant tissues is a widely used tool to investigate biochemical and physiological processes. While we have a good understanding about the hydrological cycle in plants with an evaporative enrichment in 18O in leaf water, we still lack knowledge about the biochemical link between the oxygen atoms in leaf water, leaf assimilates, and stem cellulose and associated isotope fractionations. Especially, the influence of different environmental factors on δ18O of individual carbohydrates (i.e. sugars) and thus on δ18O of cellulose is not fully resolved. A better understanding of these processes may improve climatic reconstructions of tree-ring studies about past environmental conditions. However, further progress in this topic is limited since a precise and reliable method to determine δ18O of individual sugars has not been available yet. With our new approach we attempt to overcome this issue by establishing a new methylation derivatization method suitable for GC-pyrolysis -IRMS. A methyl group (CH3) was thereby added to all hydroxyl groups of a sugar (e.g., glucose, fructose, and sucrose) during a catalyzed one-pot reaction overnight in acetonitrile with methyl iodide (CH3-I) and silver oxide, making them amenable for GC analysis. First results show a very good precision for δ18O of sucrose, but also δ18O of other high-abundant sugars such as glucose and fructose could be measured for the first time. We successfully analyzed a standard mix of all three sugars and determined various other carbohydrates not only related to plant sciences (e.g. mannitol, lactose), showing promising δ18O results. First tests with real plant samples were performed to make this method available for determining δ18O of individual carbohydrates of diverse plant tissues. In future, this new methylation derivatization method should allow us analyzing plant samples of different field sites and of lab experiments to investigate the link between leaf sugars and stem cellulose, as well as the influence of environmental factors on δ18O of individual carbohydrates in plants, but may be applied also in other research fields working on food quality or on medical applications.